



8

SEQUENCE LISTING

<110> Oxford Biomedica (UK) Limited
Kingsman, Alan
Kingsman, Susan Mary
Bebbington, Christopher Robert
Carrol, Miles William
Ellard, Fiona Margaret
Myers, Kevin Alan

<120> Antibodies

<130> 674523-2012

<140> 10/016,686

<141> 2001-11-02

<150> PCT/GB00/04317

<151> 2000-11-13

<160> 37

<170> PatentIn version 3.1

<210> 1

<211> 243

<212> PRT

<213> Artificial Sequence

<220>

<223> sequence of the mature secreted protein

<400> 1

Glu Val Gln Leu Gln Gln Ser Gly Pro Asp Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Ser Phe Thr Gly Tyr
20 25 30

Tyr Met His Trp Val Lys Gln Ser His Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly Arg Ile Asn Pro Asn Asn Gly Val Thr Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Asp Lys Ala Ile Leu Thr Val Asp Lys Ser Ser Thr Thr Ala Tyr
65 70 75 80

Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Ser Thr Met Ile Thr Asn Tyr Val Met Asp Tyr Trp Gly Gln
 100 105 110

Val Thr Ser Val Thr Val Ser Ser Gly Gly Gly Gly Ser Gly Gly Gly
 115 120 125

Gly Thr Gly Gly Gly Gly Ser Ser Ile Val Met Thr Gln Thr Pro Thr
 130 135 140

Phe Leu Leu Val Ser Ala Gly Asp Arg Val Thr Ile Thr Cys Lys Ala
 145 150 155 160

Ser Gln Ser Val Ser Asn Asp Val Ala Trp Tyr Gln Gln Lys Pro Gly
 165 170 175

Gln Ser Pro Thr Leu Leu Ile Ser Tyr Thr Ser Ser Arg Tyr Ala Gly
 180 185 190

Val Pro Asp Arg Phe Ile Gly Ser Gly Tyr Gly Thr Asp Phe Thr Phe
 195 200 205

Thr Ile Ser Thr Leu Gln Ala Glu Asp Leu Ala Val Tyr Phe Cys Gln
 210 215 220

Gln Asp Tyr Asn Ser Pro Pro Thr Phe Gly Gly Gly Thr Lys Leu Glu
 225 230 235 240

Ile Lys Arg

<210> 2
 <211> 68
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Cassette 1- Translation initiation signal and signal peptide

<400> 2
 aagcttccac catgggatgg agctgtatca tcctcttctt ggtagcaaca gctacaggtg 60
 tccactcc 68

<210> 3
 <211> 488

<212> PRT

<213> Artificial Sequence

<220>

<223> deduced amino acid sequence for the B7-1.5T4.1 fusion protein

<400> 3

Met Gly His Thr Arg Arg Gln Gly Thr Ser Pro Ser Lys Cys Pro Tyr
1 5 10 15

Leu Asn Phe Phe Gln Leu Leu Val Leu Ala Gly Leu Ser His Phe Cys
20 25 30

Ser Gly Val Ile His Val Thr Lys Glu Val Lys Glu Val Ala Thr Leu
35 40 45

Ser Cys Gly His Asn Val Ser Val Glu Glu Leu Ala Gln Thr Arg Ile
50 55 60

Tyr Trp Gln Lys Glu Lys Lys Met Val Leu Thr Met Met Ser Gly Asp
65 70 75 80

Met Asn Ile Trp Pro Glu Tyr Lys Asn Arg Thr Ile Phe Asp Ile Thr
85 90 95

Asn Asn Leu Ser Ile Val Ile Leu Ala Leu Arg Pro Ser Asp Glu Gly
100 105 110

Thr Tyr Glu Cys Val Val Leu Lys Tyr Glu Lys Asp Ala Phe Lys Arg
115 120 125

Glu His Leu Ala Glu Val Thr Leu Ser Val Lys Ala Asp Phe Pro Thr
130 135 140

Pro Ser Ile Ser Asp Phe Glu Ile Pro Thr Ser Asn Ile Arg Arg Ile
145 150 155 160

Ile Cys Ser Thr Ser Gly Gly Phe Pro Glu Pro His Leu Ser Trp Leu
165 170 175

Glu Asn Gly Glu Glu Leu Asn Ala Ile Asn Thr Thr Val Ser Gln Asp
180 185 190

Pro Glu Thr Glu Leu Tyr Ala Val Ser Ser Lys Leu Asp Phe Asn Met

195

200

205

Thr Thr Asn His Ser Phe Met Cys Leu Ile Lys Tyr Gly His Leu Arg
210 215 220

Val Asn Gln Thr Phe Asn Trp Asn Thr Thr Lys Gln Glu His Phe Pro
225 230 235 240

Asp Gly Gly Gly Gly Ser Glu Val Gln Leu Gln Gln Ser Gly Pro Asp
245 250 255

Leu Val Lys Pro Gly Ala Ser Val Lys Ile Ser Cys Lys Ala Ser Gly
260 265 270

Tyr Ser Phe Thr Gly Tyr Tyr Met His Trp Val Lys Gln Ser His Gly
275 280 285

Lys Ser Leu Glu Trp Ile Gly Arg Ile Asn Pro Asn Asn Gly Val Thr
290 295 300

Leu Tyr Asn Gln Lys Phe Lys Asp Lys Ala Ile Leu Thr Val Asp Lys
305 310 315 320

Ser Ser Thr Thr Ala Tyr Met Glu Leu Arg Ser Leu Thr Ser Glu Asp
325 330 335

Ser Ala Val Tyr Tyr Cys Ala Arg Ser Thr Met Ile Thr Asn Tyr Val
340 345 350

Met Asp Tyr Trp Gly Gln Val Thr Ser Val Thr Val Ser Ser Gly Gly
355 360 365

Gly Gly Ser Gly Gly Gly Gly Thr Gly Gly Gly Gly Ser Ser Ile Val
370 375 380

Met Thr Gln Thr Pro Thr Phe Leu Leu Val Ser Ala Gly Asp Arg Val
385 390 395 400

Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Ser Asn Asp Val Ala Trp
405 410 415

Tyr Gln Gln Lys Pro Gly Gln Ser Pro Thr Leu Leu Ile Ser Tyr Thr
420 425 430

Ser Ser Arg Tyr Ala Gly Val Pro Asp Arg Phe Ile Gly Ser Gly Tyr
435 440 445

Gly Thr Asp Phe Thr Phe Thr Ile Ser Thr Leu Gln Ala Glu Asp Leu
450 455 460

Ala Val Tyr Phe Cys Gln Gln Asp Tyr Asn Ser Pro Pro Thr Phe Gly
465 470 475 480

Gly Gly Thr Lys Leu Glu Ile Lys
485

<210> 4

<211> 592

<212> PRT

<213> Artificial Sequence

<220>

<223> deduced amino acid sequence for the Ig-5T4 fusion protein

<400> 4

Met Gly Trp Ser Cys Ile Ile Leu Phe Leu Val Ala Thr Ala Thr Gly
1 5 10 15

Val His Ser Glu Val Gln Leu Gln Gln Ser Gly Pro Asp Leu Val Lys
20 25 30

Pro Gly Ala Ser Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Ser Phe
35 40 45

Thr Gly Tyr Tyr Met His Trp Val Lys Gln Ser His Gly Lys Ser Leu
50 55 60

Glu Trp Ile Gly Arg Ile Asn Pro Asn Asn Gly Val Thr Leu Tyr Asn
65 70 75 80

Gln Lys Phe Lys Asp Lys Ala Ile Leu Thr Val Asp Lys Ser Ser Thr
85 90 95

Thr Ala Tyr Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Ser Ala Val
100 105 110

Tyr Tyr Cys Ala Arg Ser Thr Met Ile Thr Asn Tyr Val Met Asp Tyr

115 120 125
 Trp Gly Gln Val Thr Ser Val Thr Val Ser Ser Gly Gly Gly Gly Ser
 130 135 140
 Gly Gly Gly Gly Thr Gly Gly Gly Gly Ser Ser Ile Val Met Thr Gln
 145 150 155 160
 Thr Pro Thr Phe Leu Leu Val Ser Ala Gly Asp Arg Val Thr Ile Thr
 165 170 175
 Cys Lys Ala Ser Gln Ser Val Ser Asn Asp Val Ala Trp Tyr Gln Gln
 180 185 190
 Lys Pro Gly Gln Ser Pro Thr Leu Leu Ile Ser Tyr Thr Ser Ser Arg
 195 200 205
 Tyr Ala Gly Val Pro Asp Arg Phe Ile Gly Ser Gly Tyr Gly Thr Asp
 210 215 220
 Phe Thr Phe Thr Ile Ser Thr Leu Gln Ala Glu Asp Leu Ala Val Tyr
 225 230 235 240
 Phe Cys Gln Gln Asp Tyr Asn Ser Pro Pro Thr Phe Gly Gly Gly Thr
 245 250 255
 Lys Leu Glu Ile Lys Arg Ala Ser Thr Lys Gly Pro Ser Val Phe Pro
 260 265 270
 Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala Ala Leu Gly
 275 280 285
 Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp Asn
 290 295 300
 Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln
 305 310 315 320
 Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser
 325 330 335
 Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His Lys Pro Ser
 340 345 350

A1
 1

Asn Thr Lys Val Asp Lys Lys Val Glu Pro Lys Ser Cys Asp Lys Thr
355 360 365

His Thr Cys Pro Pro Cys Pro Ala Pro Glu Leu Leu Gly Gly Pro Ser
370 375 380

Val Phe Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg
385 390 395 400

Thr Pro Glu Val Thr Cys Val Val Val Asp Val Ser His Glu Asp Pro
405 410 415

Glu Val Lys Phe Asn Trp Tyr Val Asp Gly Val Glu Val His Asn Ala
420 425 430

Lys Thr Lys Pro Arg Glu Glu Gln Tyr Asn Ser Thr Tyr Arg Val Val
435 440 445

Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr
450 455 460

Lys Cys Lys Val Ser Asn Lys Ala Leu Pro Ala Pro Ile Glu Lys Thr
465 470 475 480

Ile Ser Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu
485 490 495

Pro Pro Ser Arg Asp Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys
500 505 510

Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser
515 520 525

Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp
530 535 540

Ser Asp Gly Ser Phe Phe Leu Tyr Ser Lys Leu Thr Val Asp Lys Ser
545 550 555 560

Arg Trp Gln Gln Gly Asn Val Phe Ser Cys Ser Val Met His Glu Ala
565 570 575

Leu His Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Pro Gly Lys
 580 585 590

<210> 5
 <211> 729
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> DNA sequence encoding a 5T4 ScFv designated 5T4ScFv.1

<400> 5
 gaggtccagc ttcagcagtc tggacctgac ctggtgaagc ctggggcttc agtgaagata 60
 tcctgcaagg cttctgggta ctcatcact ggctactaca tgcactgggt gaagcagagc 120
 catggaaaga gccttgagtg gattggacgt attaatccta acaatgggtg tactctctac 180
 aaccagaaat tcaaggacaa ggccatatta actgtagaca agtcatccac cacagcctac 240
 atggagctcc gcagcctgac atctgaggac tctgcggtct attactgtgc aagatctact 300
 atgattacga actatgttat ggactactgg ggtcaagtaa cctcagtcac cgtctcctca 360
 ggtggtggtg ggagcgggtg tggcggcact ggcggcggcg gatctagtat tgtgatgacc 420
 cagactccca cattcctgct tgtttcagca ggagacaggg ttaccataac ctgcaaggcc 480
 agtcagagtg tgagtaatga tgtagdttg taccacaga agccagggca gtctcctaca 540
 ctgctcatat cctatacatc cagtcgctac gctggagtcc ctgacgctt cattggcagt 600
 ggatatggga cggatttcac tttcaccatc agcactttgc aggctgaaga cctggcagtt 660
 tatttctgtc agcaagatta taattctcct ccgacgttcg gtggaggcac caagctggaa 720
 atcaaacgg 729

<210> 6
 <211> 43
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> oligonucleotide used to construct flexible linker to join the extracellular domain of B7.1 and ScFv

<400> 6
 gggggtggtg ggagcgggtg tggcggcagt ggcggcggcg gaa 43

<210> 7
 <211> 1467
 <212> DNA

<213> Artificial Sequence

<220>

<223> DNA sequence encoding a B7-1.5T4.1 fusion protein

<400> 7

atggggccaca cacggaggca gggaacatca ccatccaagt gtccatacct caatttcctt 60
cagctcttgg tgctggctgg tctttctcac ttctgttcag gtgttatcca cgtgaccaag 120
gaagtgaaag aagtggcaac gctgtcctgt ggtcacaatg tttctgttga agagctggca 180
caaactcgca tctactggca aaaggagaag aaaatgggtgc tgactatgat gtctggggac 240
atgaatatat ggcccgagta caagaaccgg accatctttg atatcactaa taacctctcc 300
attgtgatcc tggtctcgcg cccatctgac gagggcacat acgagtgtgt tgttctgaag 360
tatgaaaaag acgctttcaa gcgggaacac ctggctgaag tgacgttata agtcaaagct 420
gacttcctta cacctagtat atctgacttt gaaattccaa cttctaatat tagaaggata 480
atttgctcaa cctctggagg ttttccagag cctcacctct cctgggttga aaatggagaa 540
gaattaaatg ccatcaacac aacagtttcc caagatcctg aaactgagct ctatgctgtt 600
agcagcaaac tggatttcaa tatgacaacc aaccacagct tcatgtgtct catcaagtat 660
ggacatttaa gagtgaatca gaccttcaac tgggaatacaa ccaagcaaga gcattttcct 720
gatggaggcg ggggatccga ggtccagctt cagcagtctg gacctgacct ggtgaagcct 780
ggggcttcag tgaagatata ctgcaaggct tctggttact cattcactgg ctactacatg 840
cactgggtga agcagagcca tggaaagagc cttgagtgga ttggacgtat taatcctaac 900
aatggtgtta ctctctacaa ccagaaattc aaggacaagg ccatattaac tgtagacaag 960
tcatccacca cagcctacat ggagctccgc agcctgacat ctgaggactc tgcggtctat 1020
tactgtgcaa gatctactat gattacgaac tatgttatgg actactgggg tcaagtaacc 1080
tcagtcaccg tctcctcagg tgggtggtggg agcgggtggtg gcggcactgg cggcggcgga 1140
tctagtattg tgatgacca gactcccaca ttctgcttg tttcagcagg agacagggtt 1200
accataacct gcaaggccag tcagagtgtg agtaatgatg tagcttggtta ccaacagaag 1260
ccagggcagt ctctacact gctcatatcc tatacatcca gtcgctacgc tggagtcctt 1320
gatcgcttca ttggcagtgg atatgggacg gatttcactt tcaccatcag cactttgcag 1380
gctgaagacc tggcagttta tttctgtcag caagattata attctcctcc gacgttcggt 1440
ggaggcacca agctggaaat caaataa 1467

<210> 8
<211> 1796
<212> DNA
<213> Artificial Sequence

<220>
<223> DNA sequence encoding a Ig-5T4 fusion protein

<400> 8
ctcgagccac catgggatgg agctgtatca tcctcttctt ggtagcaaca gctacaggtg 60
tccactccga ggtccagctg cagcagtctg gacctgacct ggtgaagcct ggggcttcag 120
tgaagatata ctgcaaggct tctgggttact cattcactgg ctactacatg cactgggtga 180
agcagagcca tggaaagagc cttgagtggg ttggacgtat taatcctaac aatgggtgta 240
ctctctacaa ccagaaattc aaggacaagg ccatattaac tgtagacaag tcatccacca 300
cagcctacat ggagctccgc agcctgacat ctgaggactc tgcggtctat tactgtgcaa 360
gatctactat gattacgaac tatgttatgg actactgggg tcaagtaact tcagtcaccg 420
tctcttcagg tgggtggtgg agcgggtggg gcggcactgg cggcggcgga tctagtattg 480
tgatgacca gactcccaca ttctgcttg tttcagcagg agacagggtt accataacct 540
gcaaggccag tcagagtgtg agtaatgatg tagcttggtg ccaacagaag ccagggcagt 600
ctctacact gctcatatcc tatacatcca gtcgctacgc tggagtccct gatcgcttca 660
ttggcagtgg atatgggacg gatttcactt tcaccatcag cactttgcag gctgaagacc 720
tggcagttta tttctgtcag caagattata attctcctcc gacgttcggt ggaggcacca 780
agcttgaaat caaacggggc tccaccaagg gcccatcggt cttccccctg gcacctcct 840
ccaagagcac ctctgggggc acagcggccc tgggctgcct ggtcaaggac tacttccccg 900
aaccggtgac ggtgtcgtgg aactcaggcg cctgaccag cggcgtgcac accttccccg 960
ctgtcctaca gtctcagga ctctactccc tcagcagcgt ggtgaccgtg ccctccagca 1020
gcttggggcac ccagacctac atctgcaacg tgaatcacia gccagcaac accaagggtg 1080
acaagaaagt tgagcccaa tcttgtgaca aaactcacac atgcccaccg tgcccagcac 1140
ctgaactcct ggggggaccg tcagtcttcc tcttcccccc aaaacccaag gacacctca 1200
tgatctcccg gaccttgag gtcacatgcg tgggtggtgga cgtgagccac gaagacctg 1260
aggtcaagtt caactggtac gtggacggcg tggaggtgca taatgccaa acaaagccgc 1320
gggaggagca gtacaacagc acgtaccgtg tggtcagcgt cctcaccgtc ctgcaccagg 1380
actggctgaa tggcaaggag tacaagtga aggtctcaa caaagccctc ccagcccca 1440

tcgagaaaac catctccaaa gccaaagggc agcccccgaga accacaggtg tacaccctgc 1500
 ccccatcccc ggatgagctg accaagaacc aggtcagcct gacctgcctg gtcaaaggct 1560
 tctatcccag cgacatcgcc gtggagtggg agagcaatgg gcagccggag aacaactaca 1620
 agaccacgcc tcccgtgctg gactccgacg gctccttctt cctctatagc aagctcaccg 1680
 tggacaagag caggtggcag caggggaacg tcttctcatg ctccgtgatg catgaggctc 1740
 tgcacaacca ctacacgcag aagagcctct ccctgtcccc gggtaaata ctcgag 1796

<210> 9
 <211> 738
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> DNA sequence encoding a B7-2.5T4.1 fusion protein

<400> 9
 atgggactga gtaacattct ctttgtgatg gccttctctg tctctgggtg tgctcctctg 60
 aagattcaag cttatttcaa tgagactgca gacctgccat gccaatgtgc aaactctcaa 120
 aaccaaagcc tgagtgaagt agtagtattt tggcaggacc aggaaaactt ggttctgaat 180
 gaggtatact taggcaaaga gaaatttgac agtggttcatt ccaagtatat gggccgcaca 240
 agttttgatt cggacagttg gacctgaga cttcacaatc ttcagatcaa ggacaagggc 300
 ttgtatcaat gtatcatcca tcacaaaaag cccacaggaa tgattcgcat ccaccagatg 360
 aattctgaac tgtcagtgtc tgctaacttc agtcaacctg aaatagtacc aatttctaata 420
 ataacagaaa atgtgtacat aaatttgacc tgctcatcta tacacgggta ccagaaacct 480
 aagaagatga gtgttttgct aagaaccaag aattcaacta tcgagtatga tggattatga 540
 cagaaatctc aagataatgt cacagaactg tacgacgttt ccatcagctt gtctgtttca 600
 ttccctgatg ttacgagcaa tatgaccatc ttctgtattc tggaaactga caagacgcgg 660
 cttttatctt cacctttctc tatagagctt gaggaccctc agcctcccc agaccacatt 720
 cctggaggcg ggggatcc 738

<210> 10
 <211> 246
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> deduced amino acid sequence for the B7-2.5T4.1 fusion protein

<400> 10

Met Gly Leu Ser Asn Ile Leu Phe Val Met Ala Phe Leu Leu Ser Gly
1 5 10 15

Ala Ala Pro Leu Lys Ile Gln Ala Tyr Phe Asn Glu Thr Ala Asp Leu
20 25 30

Pro Cys Gln Phe Ala Asn Ser Gln Asn Gln Ser Leu Ser Glu Leu Val
35 40 45

Val Phe Trp Gln Asp Gln Glu Asn Leu Val Leu Asn Glu Val Tyr Leu
50 55 60

Gly Lys Glu Lys Phe Asp Ser Val His Ser Lys Tyr Met Gly Arg Thr
65 70 75 80

Ser Phe Asp Ser Asp Ser Trp Thr Leu Arg Leu His Asn Leu Gln Ile
85 90 95

Lys Asp Lys Gly Leu Tyr Gln Cys Ile Ile His His Lys Lys Pro Thr
100 105 110

Gly Met Ile Arg Ile His Gln Met Asn Ser Glu Leu Ser Val Leu Ala
115 120 125

Asn Phe Ser Gln Pro Glu Ile Val Pro Ile Ser Asn Ile Thr Glu Asn
130 135 140

Val Tyr Ile Asn Leu Thr Cys Ser Ser Ile His Gly Tyr Pro Glu Pro
145 150 155 160

Lys Lys Met Ser Val Leu Leu Arg Thr Lys Asn Ser Thr Ile Glu Tyr
165 170 175

Asp Gly Ile Met Gln Lys Ser Gln Asp Asn Val Thr Glu Leu Tyr Asp
180 185 190

Val Ser Ile Ser Leu Ser Val Ser Phe Pro Asp Val Thr Ser Asn Met
195 200 205

Thr Ile Phe Cys Ile Leu Glu Thr Asp Lys Thr Arg Leu Leu Ser Ser
210 215 220

Pro Phe Ser Ile Glu Leu Glu Asp Pro Gln Pro Pro Pro Asp His Ile
225 230 235 240

Pro Gly Gly Gly Gly Ser
245

<210> 11
<211> 1518
<212> DNA
<213> Artificial Sequence

<220>
<223> B7 link ScFv sequence

<400> 11
atggccttgca attgtcagtt gatgcaggat acaccactcc tcaagtttcc atgtccaagg 60
ctcattcttc tctttgtgct gctgattcgt ctttcacaag tgtcttcaga tgttgatgaa 120
caactgtcca agtcagtgaag agataaggta ttgctgcctt gccgttataa ctctccgcat 180
gaagatgagt ctgaagaccg aatctactgg caaaaacatg acaaagtggg gctgtctgtc 240
attgctggga aactaaaagt gtggcccag tataagaacc ggactttata tgacaacact 300
acctactctc ttatcatcct gggcctgggc ctttcagacc ggggcacata cagctgtgtc 360
gttcaaaaga aggaaagagg aacgtatgaa gttaaactact tggctttagt aaagttgtcc 420
atcaaagctg acttctctac cccaacata actgagtctg gaaacccatc tgcagacact 480
aaaaggatta cctgctttgc ttccgggggt ttcccaaagc ctgcttctc ttgggtggaa 540
aatggaagag aattacctgg catcaatacg acaatttccc aggatcctga atctgaattg 600
tacaccatta gtagccaact agatttcaat acgactcgca accacaccat taagtgtctc 660
attaaatatg gagatgctca cgtgtcagag gacttcacct gggaaaaacc ccagaagac 720
cctcctgata gcaagcccgg ggggtggggg agcgggtggg gcggcagtg gcggcggcga 780
actagtgagg tccagcttca gcagtctgga cctgacctgg tgaagcctgg ggcttcagtg 840
aagatatcct gcaaggcttc tgggtactca ttactggct actacatgca ctgggtgaag 900
cagagccatg gaaagagcct tgagtggatt ggacgtatta atcctaaca tgggtgttact 960
ctctacaacc agaaattcaa ggacaaggcc atattaactg tagacaagtc atccaccaca 1020
gcctacatgg agctccgag cctgacatct gaggactctg cggcttatta ctgtgcaaga 1080
tctactatga ttacgaacta tggtatggac tactgggggc aagtaacttc agtcaccgtc 1140
tcttcagggtg gtgggtgggag cgggtggggc ggcactggcg gcggcggatc tagtattgtg 1200

atgacccaga	ctcccacatt	cctgcttggt	tcagcaggag	acagggttac	cataacctgc	1260
aaggccagtc	agagtgtgag	taatgatgta	gcttggtacc	aacagaagcc	agggcagtct	1320
cctacactgc	tcatactcta	tacatccagt	cgctacgctg	gagtccctga	tcgcttcatt	1380
ggcagtggat	atgggacgga	tttcactttc	accatcagca	ctttgcaggc	tgaagacctg	1440
gcagtttatt	tctgtcagca	agattataat	tctcctccga	cgttcgggtg	aggcaccaag	1500
ctggaaatca	aacggtaa					1518

<210> 12
 <211> 2090
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> ScFv-IgE

<400> 12	
ctcgagccac	catgggatgg agctgtatca tcctcttctt ggtagcaaca gctacaggtg 60
tccactccga	ggtccagctg cagcagtctg gacctgacct ggtgaagcct ggggcttcag 120
tgaagatata	ctgcaaggct tctgggttact cattcactgg ctactacatg cactgggtga 180
agcagagcca	tggaaagagc cttgagtggg ttggacgtat taatcctaac aatggtgtta 240
ctctctacaa	ccagaaattc aaggacaagg ccatattaac tgtagacaag tcatccacca 300
cagcctacat	ggagctccgc agcctgacat ctgaggactc tgcggtctat tactgtgcaa 360
gatctactat	gattacgaac tatgttatgg actactgggg tcaagtaact tcagtcaccg 420
tctcttcagg	tggtgggtggg agcgggtggg ggggactgg cggcggcgga tctagtattg 480
tgatgacca	gactcccaca ttctgcttg tttcagcagg agacagggtt accataacct 540
gcaaggccag	tcagagtgtg agtaatgatg tagcttggtg ccaacagaag ccagggcagt 600
ctcctacact	gctcatatcc tatacatcca gtcgctacgc tggagtcctt gatcgcttca 660
ttggcagtgg	atatgggacg gatttcactt tcaccatcag cactttgcag gctgaagacc 720
tggcagttta	tttctgtcag caagattata attctctctc gacgttcggt ggaggcacca 780
agcttgaaat	caaacggggc tccacacaga gccatccgt cttccccttg acccgctgct 840
gcaaaaacat	tccctccaat gccacctccg tgactctggg ctgcctggcc acgggctact 900
tcccggagcc	ggtgatgggtg acctgggaca caggctccct caacgggaca actatgacct 960
taccagccac	caccctcacg ctctctgggc actatgccac catcagcttg ctgaccgtct 1020

AI

cggggtgcgtg ggccaagcag atgttcacct gccgtgtggc acacactcca tcgtccacag 1080
 actgggtcga caacaaaacc ttcagcgtct gctccaggga cttcaccccg cccaccgtga 1140
 agatcttaca gtcgtcctgc gacggcgggc ggcacttccc cccgaccatc cagctcctgt 1200
 gcctcgtctc tgggtacacc ccagggacta tcaacatcac ctggctggag gacgggcagg 1260
 tcatggacgt ggacttgtcc accgcctcta ccacgcagga gggtagctg gcctccacac 1320
 aaagcgagct caccctcagc cagaagcact ggctgtcaga ccgcacctac acctgccagg 1380
 tcacctatca aggtcacacc tttgaggaca gcaccaagaa gtgtgcagat tccaaccgga 1440
 gaggggtgag cgcctaccta agccggccca gcccggttca cctgttcac cgcgaagtgc 1500
 ccacgatcac ctgtctggtg gtggacctgg caccagcaa ggggaccgtg aacctgacct 1560
 ggtcccgggc cagtgggaag cctgtgaacc actccaccag aaaggaggag aagcagcgca 1620
 atggcacgtt aaccgtcacg tccaccctgc cgggtggcac ccgagactgg atcgaggggg 1680
 agacctacca gtgcaggggtg acccaccccc acctgccag ggcctcatg cgggtccacga 1740
 ccaagaccag cggcccgct gctgccccgg aagtctatgc gtttgcgacg ccggagtggc 1800
 cggggagccg ggacaagcgc accctcgcct gcctgatcca gaacttcatg cctgaggaca 1860
 tctcgggtgca gtggctgcac aacgaggtgc agtccccga cggccggcac agcacgacgc 1920
 agccccgcaa gaccaagggc tccggcttct tcgtcttcag ccgcctggag gtgaccaggg 1980
 ccgaatggga gcagaaagat gagttcatct gccgtgcagt ccatgaggga gcgagccct 2040
 cacagaccgt ccagcgagcg gtgtctgtaa atcccggtaa atgagagctc 2090

<210> 13
 <211> 945
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> B7-EGF

<400> 13
 atggcttgca attgtcagtt gatgcaggat acaccactcc tcaagtttcc atgtccaagg 60
 ctcattcttc tctttgtgct gctgattcgt ctttcacaag tgtcttcaga tgttgatgaa 120
 caactgtcca agtcagtga agataaggta ttgtgcctt gccgttaca ctctccgat 180
 gaagatgagt ctgaagaccg aatctactgg caaaaacatg acaaagtggg gctgtctgtc 240
 attgctggga aactaaaagt gtggccccgag tataagaacc ggactttata tgacaacact 300
 acctactctc ttatcatcct gggcctggtc ctttcagacc ggggcacata cagctgtgtc 360

gttcaaaaga aggaaagagg aacgtatgaa gttaaact tggctttagt aaagttgtcc 420
atcaaagctg acttctctac cccaacata actgagtctg gaaacccatc tgcagacact 480
aaaaggatta cctgctttgc ttccgggggt ttcccaaagc ctgctttctc ttggttgga 540
aatggaagag aattacctgg catcaatagc acaatttccc aggatcctga atctgaattg 600
tacaccatta gtagccaact agatttcaat acgactcgca accacacat taagtgtctc 660
attaaatatg gagatgctca cgtgtcagag gacttcacct gggaaaaacc ccagaagac 720
cctcctgata gcaagcccgg ggggtggtggg agcgggtggtg gcggcagtg gcggcggcga 780
actagtaata gtgactctga atgtcccctg tcccacgatg ggtactgcct ccatgatggt 840
gtgtgcatgt atattgaagc attggacaag tatgcatgca actgtgttgt tggctacatc 900
ggggagcgat gtcagtaccg agacctgaag tgggtgggaac tgcgc 945

<210> 14
<211> 1263
<212> DNA
<213> Artificial Sequence

<220>
<223> canine 5T4 polypeptide having the amino acid sequence

AI
<400> 14
atgcctgggg ggtgctccc gggccccgcc gccggggacg ggcgggtgcg gctggcgcg 60
ctggcgctgg tgctcctggg ctgggtctcc tegtctctgc tcacctctg ggcgcctcc 120
gccgccgctt ccacgtcgcc gccggcctcc gcggcgctcg ccccgcccc gctgccgggc 180
cagtgcctcc agccttgca gtgctcggag gcggcgcgca cggtaagtg cgttaaccgc 240
aacctgaccg aggtgcccgc ggacctgcc cctacgtgc gcaacctctt cctcacgggc 300
aaccagctgg cgggtgtgcc ccccggcgc ttccgccgc ggcgcgcgt ggccgagctg 360
gccgcgtca acctgagcgg cagcagcctg cgggaggtgt gcgccggcgc cttcgagcac 420
ctgccagcc tgcgccagct cgacctcagc cacaaccgc tgggcaacct cagcgcttc 480
gccttcgcgg gcagcgacgc cagccgctcg ggccccagcc cctggtgga gctgatgctg 540
aaccacatcg tgcccccgga cgaccggcgg cagaaccgga gcttcgagg catggtggcg 600
gctgcctcc gagcggggcg cgcgcttcgc gggctgcagt gcctggagct ggccggcaac 660
cgcttctct acttgctcg cgacgtcctg gccagctac ccggcctcc gcacctggac 720
ctgcgcaaca actcctggt gagcctcacc tacgtgtcct tccgcaacct gacgcacttg 780

gagagcctcc acctggagga caacgccctc aaggtccttc acaacgccac cctggcggag 840
ctgcagagcc tgccccacgt ccgggtcttc ctggacaaca acccctgggt ctgcgattgt 900
cacatggcag acatggtggc ctggctcaag gagacagagg tggtgccggg caaagccggg 960
ctcacctgtg cattccccga gaaaatgagg aatcgggccc tcttggaact caacagctcc 1020
cacctggact gtgaccctat cctccctcca tccctgcaga cttcttatgt cttcctaggt 1080
attgtcttag ccctgatagg cgccatcttc ctactggttt tgtatttgaa ccgcaagggg 1140
ataaagaagt ggatgcataa catcagagat gcctgcaggg atcacatgga agggatatcac 1200
tacagatacg aaatcaatgc agaccccagg ttaacaaacc tcagttccaa ttcggatgtc 1260
tga 1263

<210> 15
<211> 420
<212> PRT
<213> Artificial Sequence

<220>
<223> canine 5T4 polypeptide having the amino acid sequence
<400> 15

Met Pro Gly Gly Cys Ser Arg Gly Pro Ala Ala Gly Asp Gly Arg Leu
1 5 10 15

Arg Leu Ala Arg Leu Ala Leu Val Leu Leu Gly Trp Val Ser Ser Ser
20 25 30

Ser Leu Thr Ser Trp Ala Pro Ser Ala Ala Ala Ser Thr Ser Pro Pro
35 40 45

Ala Ser Ala Ala Ser Ala Pro Pro Pro Leu Pro Gly Gln Cys Pro Gln
50 55 60

Pro Cys Glu Cys Ser Glu Ala Ala Arg Thr Val Lys Cys Val Asn Arg
65 70 75 80

Asn Leu Thr Glu Val Pro Ala Asp Leu Pro Pro Tyr Val Arg Asn Leu
85 90 95

Phe Leu Thr Gly Asn Gln Leu Ala Val Leu Pro Pro Gly Ala Phe Ala
100 105 110

Al
d

Arg Arg Pro Pro Leu Ala Glu Leu Ala Ala Leu Asn Leu Ser Gly Ser
115 120 125

Ser Leu Arg Glu Val Cys Ala Gly Ala Phe Glu His Leu Pro Ser Leu
130 135 140

Arg Gln Leu Asp Leu Ser His Asn Pro Leu Gly Asn Leu Ser Ala Phe
145 150 155 160

Ala Phe Ala Gly Ser Asp Ala Ser Arg Ser Gly Pro Ser Pro Leu Val
165 170 175

Glu Leu Met Leu Asn His Ile Val Pro Pro Asp Asp Arg Arg Gln Asn
180 185 190

Arg Ser Phe Glu Gly Met Val Ala Ala Ala Leu Arg Ala Gly Arg Ala
195 200 205

Leu Arg Gly Leu Gln Cys Leu Glu Leu Ala Gly Asn Arg Phe Leu Tyr
210 215 220

Leu Pro Arg Asp Val Leu Ala Gln Leu Pro Gly Leu Arg His Leu Asp
225 230 235 240

Leu Arg Asn Asn Ser Leu Val Ser Leu Thr Tyr Val Ser Phe Arg Asn
245 250 255

Leu Thr His Leu Glu Ser Leu His Leu Glu Asp Asn Ala Leu Lys Val
260 265 270

Leu His Asn Ala Thr Leu Ala Glu Leu Gln Ser Leu Pro His Val Arg
275 280 285

Val Phe Leu Asp Asn Asn Pro Trp Val Cys Asp Cys His Met Ala Asp
290 295 300

Met Val Ala Trp Leu Lys Glu Thr Glu Val Val Pro Gly Lys Ala Gly
305 310 315 320

Leu Thr Cys Ala Phe Pro Glu Lys Met Arg Asn Arg Ala Leu Leu Glu
325 330 335

Leu Asn Ser Ser His Leu Asp Cys Asp Pro Ile Leu Pro Pro Ser Leu

Al
4

340

345

350

Gln Thr Ser Tyr Val Phe Leu Gly Ile Val Leu Ala Leu Ile Gly Ala
 355 360 365

Ile Phe Leu Leu Val Leu Tyr Leu Asn Arg Lys Gly Ile Lys Lys Trp
 370 375 380

Met His Asn Ile Arg Asp Ala Cys Arg Asp His Met Glu Gly Tyr His
 385 390 395 400

Tyr Arg Tyr Glu Ile Asn Ala Asp Pro Arg Leu Thr Asn Leu Ser Ser
 405 410 415

Asn Ser Asp Val
 420

<210> 16
 <211> 47
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> oligonucleotide used to construct flexible linker to join the extracellular domain of B7.1 and ScFv

<400> 16
 ctagttccgc cgccgccact gccgccacca ccgctccac caccccc 47

<210> 17
 <211> 38
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Forward primer used in PCR reaction to introduce 5' EcoR1 and 3' Sma I sites

<400> 17
 ctcgaattcc accatggctt gcaattgtca gttgatgc 38

<210> 18
 <211> 30
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Reverse primer used in PCR reaction to introduce 5' EcoR1 and 3' Sma I sites

<400> 18
ctccccgggc ttgctatcag gagggctcttc 30

<210> 19
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> Forward primer used to amplify 5T4 specific ScFv

<400> 19
ctcactagtg aggtccagct tcagcagtc 29

<210> 20
<211> 44
<212> DNA
<213> Artificial Sequence

<220>
<223> Reverse primer used to amplify 5T4 specific ScFv

<400> 20
ctcgcggccg cttaccgttt gatttccagc ttggtgcctc cacc 44

Al
1
<210> 21
<211> 87
<212> DNA
<213> Artificial sequence

<220>
<223> complementary single stranded oligonucleotide encoding a translation initiation sequence and the human immunoglobulin kappa a light chain signal peptide

<400> 21
ctagactcga gccaccatgg gatggagctg tatcatcctc ttcttggtag caacagctac 60

aggtgtccac tccgaggtcc agctgca 87

<210> 22
<211> 79
<212> DNA
<213> Artificial Sequence

<220>
<223> complementary single stranded oligonucleotide encoding a translation initiation sequence and the human immunoglobulin kappa a light chain signal peptide

<400> 22
gctggacctc ggagtggaca cctgtagctg ttgctaccaa gaagaggatg atacagctcc 60

atcccatggt ggctcgagt

79

<210> 23
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify 5T4 ScFv

<400> 23
gtccagctgc agcagtctgg

20

<210> 24
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify 5T4 ScFv

<400> 24
cgtttgattt caagcttggt gc

22

<210> 25
<211> 40
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify HIgG1 constant region

<400> 25
gcgcaagctt gaaatcaaac gggcctccac caagggccca

40

<210> 26
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify HIgG1 constant region

<400> 26
gcgctcgag tcatttaccg ggagacaggg

30

<210> 27
<211> 40
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify fusion construct

<400> 27
gcgcaagctt gaaatcaaac gggcctccac acagagccca 40

<210> 28
<211> 31
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify fusion construct

<400> 28
gcgctcgag tcatttaccg ggatttacag a 31

<210> 29
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify DNA

<400> 29
ggactagtaa tagtgactct gaatgtccc 29

AI
<210> 30
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> PCR primer used to amplify DNA

<400> 30
attagcggcc gcttagcgca gttcccacca cttc 34

<210> 31
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> B7-Sbf primer for B7-5T4 scFv

<400> 31
atcgctgca ggccaccatg gcttgcaatt gtcag 35

<210> 32
<211> 34

<212> DNA
<213> Artificial Sequence

<220>
<223> 5T4sc-RI primer for B7-5T4 scFv

<400> 32
gcgcgaattc ttaccgtttg atttccagct tggt

34

<210> 33
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> L-Sbf primer for L-5T4scFv

<400> 33
atcgctgca ggccaccatg ggatggagct gtat

34

<210> 34
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> 5T4sc-RI primer for L-5T4scFv

<400> 34
gcgcgaattc ttaccgtttg atttccagct tggt

34

<210> 35
<211> 48
<212> DNA
<213> Artificial Sequence

<220>
<223> L-Sbf primer used to prepare L-5T4 scFv

<400> 35
ctagtaccgg tgggtggtggg agcgggtggtg gcggcagtg cggcggcg

48

<210> 36
<211> 15
<212> PRT
<213> Artificial Sequence

<220>
<223> 5T4sc-RI primer used to prepare L-5T4 scFv

<400> 36

Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser

1

5

10

15

<210> 37

<211> 76

<212> DNA

<213> Artificial Sequence

<220>

<223> Leader sequence in pBS II

<400> 37

ctagacctgc aggccaccat gggatggagc tgtatcatcc tcttcttggt agcaacagct 60

acaggtgtac actccc 76

A1

✓